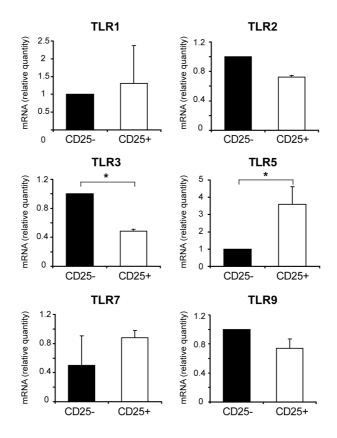


Supplementary 1

TLR profile of CD4+ T cells in comparison with IL-10-Treg.

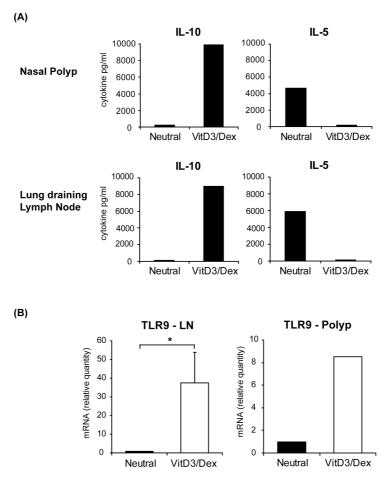
Freshly isolated CD4+ T cells and day 14 IL-10-Tregs (VitD3/Dex-Tregs) from the same donor were assessed for expression of TLR transcripts by Real time RT-PCR. Data are shown normalised to an endogenous control (18s rRNA) and expressed relative to day 0 CD4+ T cells. A representative experiment of a minimum of 4 performed using different healthy donors is shown.



Supplementary 2

Complete TLR mRNA profile of human CD4+CD25+ Treg.

CD4+ CD25+/- T cells were isolated by cell sorting on the basis of high or negative expression of CD25, respectively. Purity of both populations was routinely >99%. The TLR expression profile was analysed by Real time RT-PCR. Data are shown normalised to an endogenous control (18s rRNA) and expressed relative to CD25- T cells. Mean data ± SEM from 3 healthy donors are depicted. \*=p<0.05, as assessed by Mann Whitney rank sum test.

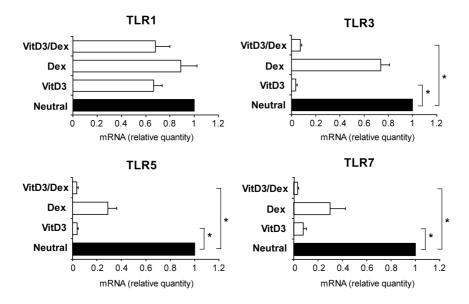


Supplementary 3

Drug-induced IL-10-Treg can be induced from T cells derived from human respiratory tract tissue and express TLR9.

CD3+CD4+ T cells and CD3+ T cells were isolated from human lung draining lymph nodes and nasal polyps, respectively and then cultured for two 7-day cycles with anti-CD3, IL-2 and IL-4 alone (Neutral) or additionally with 1α25dihydroxyvitamin D3 (VitD3) and dexamethasone (Dex).

- (A) Cells were re-stimulated for 48 hours with anti-CD3 and IL-2 alone and the supernatants were analysed for expression of IL-10 and IL-5 by ELISA. Data shown are representative of 3 independent experiments.
- (B) Gene expression of TLR9 at day 14 as assessed by Real time RT-PCR. Data are shown normalised to an endogenous control (18s rRNA) and expressed relative to Neutral cells. Mean data ± SEM from 3 lymph node (LN) samples are depicted. Data are representative of 2 independent nasal polyp experiments. \*=p<0.05 as assessed by Mann Whitney rank sum test.



## **Supplementary 4**

The effects of  $1\alpha$ ,25-dihydroxyvitamin D3 (VitD3) and dexamethasone (Dex), singly or in combination, on the expression of TLR by CD4+ T cells at day 14, as examined by Real time RT-PCR. Data are shown normalised to an endogenous control (18s rRNA) and expressed relative to Neutral cells. Mean data  $\pm$  standard error of the mean from 5 healthy donors are presented. \*=p<0.05 as assessed by One Way ANOVA on Ranks.